

71007

Attachment H

COVER SHEET (PAGE 1 of 2)

May 1998 CALFED ECOSYSTEM RESTORATION PROPOSAL SOLICITATION

Proposal Title: Biological Assessment of Green Sturgeon in the Sacramento-San Joaquin Watershed
Applicant Name: Joseph J. Cech, Jr., Ph.D.
Mailing Address: Dept. Wildlife, Fish, & Conservation Biology, UC Davis, Davis, CA 95616
Telephone: (530) 752-3103
Fax: (530) 752-4154

Amount of funding requested: \$ 397,742 for 2 years

Indicate the Topic for which you are applying (check only one box). Note that this is an important decision: see page of the Proposal Solicitation Package for more information.

- | | |
|---|--|
| <input type="checkbox"/> Fish Passage Assessment | <input type="checkbox"/> Fish Passage Improvements |
| <input type="checkbox"/> Floodplain and Habitat Restoration | <input type="checkbox"/> Gravel Restoration |
| <input type="checkbox"/> Fish Harvest | <input checked="" type="checkbox"/> Species Life History Studies |
| <input type="checkbox"/> Watershed Planning/Implementation | <input type="checkbox"/> Education |
| <input type="checkbox"/> Fish Screen Evaluations - Alternatives and Biological Priorities | |

Indicate the geographic area of your proposal (check only one box):

- | | |
|--|---|
| <input type="checkbox"/> Sacramento River Mainstem | <input type="checkbox"/> Sacramento Tributary: <u> </u> |
| <input checked="" type="checkbox"/> Delta (incl. Feather R. watershed) | <input type="checkbox"/> East Side Delta Tributary: <u> </u> |
| <input type="checkbox"/> Suisun Marsh and Bay | <input type="checkbox"/> San Joaquin Tributary: <u> </u> |
| <input type="checkbox"/> San Joaquin River Mainstem | <input type="checkbox"/> Other: <u> </u> |
| <input type="checkbox"/> Landscape (entire Bay-Delta watershed) | <input type="checkbox"/> North Bay: <u> </u> |

Indicate the primary species which the proposal addresses (check no more than two boxes):

- | | |
|--|--|
| <input type="checkbox"/> San Joaquin and East-side Delta tributaries fall-run chinook salmon | |
| <input type="checkbox"/> Winter-run chinook salmon | <input type="checkbox"/> Spring-run chinook salmon |
| <input type="checkbox"/> Late-fall run chinook salmon | <input type="checkbox"/> Fall-run chinook salmon |
| <input type="checkbox"/> Delta smelt | <input type="checkbox"/> Longfin smelt |
| <input type="checkbox"/> Splittail | <input type="checkbox"/> Steelhead trout |
| <input checked="" type="checkbox"/> Green sturgeon | <input type="checkbox"/> Striped bass |
| <input type="checkbox"/> Migratory birds | |

COVER SHEET (PAGE 2 of 2)

May 1998 CALFED ECOSYSTEM RESTORATION PROPOSAL SOLICITATION

Indicate the type of applicant (check only one box):

- | | |
|--|---|
| <input type="checkbox"/> State agency | <input type="checkbox"/> Federal agency |
| <input type="checkbox"/> Public/Non-profit joint venture | <input type="checkbox"/> Non-profit |
| <input type="checkbox"/> Local government/district | <input type="checkbox"/> Private party |
| <input checked="" type="checkbox"/> University | <input type="checkbox"/> Other: _____ |

Indicate the type of project (check only one box):

- | | |
|--|---|
| <input type="checkbox"/> Planning | <input type="checkbox"/> Implementation |
| <input type="checkbox"/> Monitoring | <input type="checkbox"/> Education |
| <input checked="" type="checkbox"/> Research | |

By signing below, the applicant declares the following:

- (1) the truthfulness of all representations in their proposal;
- (2) the individual signing the form is entitled to submit the application on behalf of the applicant (if applicant is an entity or organization); and
- (3) the person submitting the application has read and understood the conflict of interest and confidentiality discussion in the PSP (Section II.K) and waives any and all rights to privacy and confidentiality of the proposal on behalf of the applicant, to the extent as provided in the Section.

Joseph A. Bohm
(Signature of Applicant)



Office of the Vice Chancellor for Research
410 Mrak Hall, One Shields Avenue
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Facsimile: 530.752.5432
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CALFED Bay-Delta Program Office
1416 Ninth Street, Suite 1155
Sacramento, CA 95814

JUN 2 9 1998

Research Proposal Entitled
Biological Assessment of Green Sturgeon in the Sacramento-San Joaquin Watershed
Principal Investigator - **Joseph J. Cech, Jr.**

Dear Colleague:

It is a pleasure to present for your consideration the referenced proposal.

It is our understanding that for purposes of determining applicant type, The Regents will be classified as "Agency" thereby resulting awards will only include the terms identified in Attachment D and Item 3 of the 1998 Proposal Solicitation Package.

The University takes exception to clauses pertaining to Substitution, Rights in Data and Indemnification as detailed in Attachment D. On behalf of the Regents of the University of California, we hereby reserve the right to negotiate said clauses as detailed in the Proposal Solicitation Package should this proposal result in a subsequent award.

Please call on the principal investigator for scientific information. Administrative questions may be directed to me or to Petrina Ho by telephone, facsimile or electronic mail at the numbers specified above. We request that correspondence pertaining to this proposal and a subsequent award be sent to the Office of Research and to the principal investigator.

Sincerely,

A handwritten signature in cursive script that reads "Sandra M. Dowdy".
Sandra M. Dowdy
Contracts and Grants Analyst

Enclosure

cc: J. J. Cech, Jr.

Proposal to:

Name CALFED Bay-Delta Program
Address 1416 Ninth St., Suite 1155, Sacramento, CA 95814

Submitting Organization:

The Regents of the University of California
University of California
Davis, California 95616

Title of Proposed Research:

Biological Assessment of Green Sturgeon in the Sacramento-San Joaquin Watershed

Total Amount Requested:

\$397,742

Proposed Duration:

2 years

Desired Starting Date:

10-1-98

Principal Investigator/

Co-Principal Investigator(s): **Department:** **Phone Number:**

Joseph J. Cech, Jr. Wildlife, Fish, & Cons. Biol. (530) 752-3103
Serge I. Doroshov, Animal Sci., (530) 752-7603; Gary P. Moberg, Animal Sci., (530)
752-0233; Bernard P. May, Animal Sci. (530) 754-8123; Raymond G. Schaffter, Cal Fish &
Game (209) 948-7081; David M. Kohlhorst, Cal. Fish & Game (209) 948-7080

Checks Made Payable to:

The Regents of the University of California

Send Checks to:

Cashier's Office
University of California
Davis, CA 95616

Send Award Notice to:

Office of Research
University of California
Davis, California 95616
(916) 752-2075

Approvals:

Joseph J. Cech, Jr. 6-18-98 [Signature] 7-2-98
Principal Investigator Date Co-Principal Investigator Date

[Signature] 6-18-98
Co-Principal Investigator Date Department Chair Date

Dean, College/School Date

Sandra M. Doroshov
Official Signing for Organization

II. Executive Summary

A. Project Title and Applicant Name: BIOLOGICAL ASSESSMENT OF GREEN STURGEON IN THE SACRAMENTO-SAN JOAQUIN WATERSHED; Joseph J. Cech, Jr.

B. Project Description and Primary Biological/Ecological Objectives: The green sturgeon (GS, *Acipenser medirostris*) is a CALFED priority (tier one) species, and the proposed work will focus on the biological characteristics of this species and its habitats towards their eventual restoration. Our coordinated UC Davis-CDFG team will address key areas of scientific uncertainty about GS to minimize harm to this species and its population(s) in the lower Sacramento-San Joaquin watershed. In the first two phases (years) of a multi-phase investigation, we will determine baseline information regarding this species' biological requirements in the Sacramento-San Joaquin watershed, and the feasibility of GS culture for future mitigational considerations. The project has five objectives: 1. determine juvenile GS' food and oxygen requirements at different environmental temperatures, temperature tolerance limits and behavioral tendencies, and swimming performance; 2. determine GS' requirements for gonadal development, spawning, and the successful rearing of larvae and fry; 3. determine potential environmental stressors' effects on GS' reproductive functioning and well being; 4. determine the genetic stock structure of naturally spawning GS from the Sacramento-San Joaquin river system; and 5. determine GS spawning site suitability in the Feather River and provide GS adults, sub-adults, larvae, and eggs from the Sacramento system to UC Davis scientists conducting the studies outlined in the first four objectives. These projects will provide valuable information to decision-makers regarding environmental resource management options to restore Bay-Delta ecological health and water quality.

C. Approach/Tasks/Schedule: We will determine the physiological/behavioral responses and limits, reproductive/early life history requirements, stress responses, genetic makeup, and spawning locations/requirements of GS in the lower Sacramento-San Joaquin watershed. We will capture juvenile GS from Feather and Sacramento Rivers (including cooperative arrangements with Curt Brown, USFWS). Sturgeon egg and larval sampling will be conducted with simultaneous temperature, depth, flow velocity and substrate composition measurements during the late winter and spring of 1999 and 2000. Sturgeon eggs will be either maintained alive or preserved and transported to UC Davis researchers. During the fall of 1998, modifications will be made to procedures of a scheduled sturgeon tagging study so that adult and sub-adult green sturgeon captured during this tagging can be held alive until they can be transported to the UC Davis Aquatic Center. During juvenile longline sampling in the west delta during August and November of 1998 and 1999, all GS juveniles will be made available for UC Davis scientists for live pickup at various delta marinas. With USFWS and tribal (Yurok, Hoopa) cooperation, we will capture a few brood fish (for captive spawning purposes) from the Klamath basin (where spawning GS are more likely captured) to provide more fish for the proposed studies. At UC Davis (Phase 1), we will determine GS' temperature tolerance limits and behavioral tendencies, swimming performance limits, developmental progress of gonadal tissues and germ cells, rhythms of circulating hormones and gonadal responses to stressors (including culture conditions), molecular markers to differentiate WS and GS and their relative proportion spawning in the Sacramento-San Joaquin

drainage, and GS genetic polymorphisms. During Phase 2, we will continue spawning location/requirements studies, determine temperature's influence on food and oxygen consumption rates, optimum artificial spawning techniques, and degree of reproductive isolation of Sacramento-San Joaquin GS from Klamath River and Rogue River (Oregon) populations. Phase 3 studies will depend upon results from the first two phases.

D. Justification for Project and Funding by CALFED: CALFED funding is proposed because of great need for species-specific information on GS in the natural environment and in laboratory and culture conditions. Because this species is rare and only lightly harvested in California, there is little justification for funding from normal sport or commercial fisheries research funding sources. Without determining the GS' vulnerability to temperature stress and flow characteristics, spawning and early rearing requirements, responses to stressful environments, and populational identity, we cannot justify flow and other recommendations for maintenance and preservation of this species.

E. Budget Costs and Third Party Impacts: Requested CALFED funding is \$199,046 for FY 1998-99 and \$198,696 for FY 1999-00. This amount includes funds for salaries and benefits of personnel (graduate students' and seasonal employees' support; and partial support of a staff research associate, CDFG investigator, and associate research scientist), student fee remissions, equipment, supplies/expenses, operation of vehicles and vessels, and overhead (@10% of non-student fee remissions and non-equipment costs). "Leveraged" support (\$96,552) will be provided by UC Davis (5% of investigators' salaries and benefits while working on the GS project), and an estimated \$30,000 of support by using CDFG personnel and vessels conducting white sturgeon (WS) research funded by Federal Sport Fisheries Restoration Funds and matching state funds for obtaining GS, and by using State Water Project facilities and personnel to collect juveniles at the Byron fish screens. Eventual enhanced sport and native Californian fisheries and more flexible water management strategies should benefit third parties.

F. Applicant Qualifications: Drs. Joseph J. Cech, Jr., Serge I. Doroshov, Gary P. Moberg, and Bernard P. May are well-known, UC Davis fish (including sturgeon) biologists; and Raymond G. Schaffter and David Kohlhorst are CDFG's recognized sturgeon authorities in the Bay-Delta.

G. Monitoring and Data Evaluation: Data collection and evaluation will follow established procedures. In addition to quarterly, annual and final reports, results will be presented at interagency workgroup meetings, workshops, and professional scientific meetings, and published in peer-reviewed journals. Feather River temperature data will be entered on the CDFG web page within 30 days of data collection.

H. Local Support/Coordination with Other Programs/Compatibility with CALFED Objectives: Most of the infrastructure/equipment required for this project is already available at UC Davis and CDFG Bay Delta and Special Water Projects Division. Assistance from USFWS (Curt Brown, Tom Kisanuki) is being arranged, and coordination with the Yurok and Hoopa Tribal Fisheries biologists has been initiated. Increased knowledge of this priority (tier one) species will potentially assist several CALFED projects.

III. Title of project:

BIOLOGICAL ASSESSMENT OF GREEN STURGEON IN THE SACRAMENTO-SAN JOAQUIN WATERSHED

Principle Investigator:

Joseph J. Cech, Jr., Professor, Department of Wildlife, Fish, and Conservation Biology, University of California, Davis, CA 95616, Phone: (530) 752-3103, FAX (530) 752-4154, jjcech@ucdavis.edu

Co-Principal Investigators:

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Gary P. Moberg, Professor and Associate Dean for Animal Biology, College of Agricultural and Environmental Sciences, Department of Animal Science, University of California, Davis, CA 95616,
Phone: (530) 752-0233, (530) 752-1253, gpmoberg@ucdavis.edu

Bernard P. May, Associate Researcher, Department of Animal Science, University of California, Davis, CA 95616, Phone: (530) 754-8123, bpmay@ucdavis.edu

Raymond G. Schaffter, Fishery Biologist, California Department of Fish and Game, Bay Delta and Special Water Projects Division, 4001 N. Wilson Way, Stockton CA 95205, Phone: (209) 948-7081, FAX (209) 946-6355, rschafft@delta.dfg.ca.gov

David M. Kohlhorst, Senior Fishery Biologist, California Department of Fish and Game, Bay Delta and Special Water Projects Division, 4001 N. Wilson Way, Stockton CA 95205, Phone: (209) 948-7080, FAX (209) 946-6355, dkohlhor@delta.dfg.ca.gov

Type of Organization and Tax Status:

UC Davis is a State-assisted public research and educational institution. California Department of Fish and Game is a Constitutionally mandated agency of the State of California. (Non-Profit, exempt under status 501(c)(3) of the IRS code of 1954 under Type of organization and Tax Status)

Tax Identification Number: 94-603-6494 (UC Davis)

Participants and Collaborators in Implementation:

US Fish and Wildlife Service
Yurok and Hoopa Tribes (pending)

IV. PROJECT DESCRIPTION

a. Project Description and Approach:

1. Objectives: The project has five objectives that combine agency and university biological expertise and link laboratory and field approaches: 1. determine juvenile green sturgeon's (GS) food and oxygen requirements at different environmental temperatures, temperature tolerance limits and behavioral tendencies, and swimming performance; 2. determine GS' requirements for gonadal development, spawning, and the successful rearing of larvae and fry; 3. determine potential environmental stressors' effects on GS' reproductive functioning and well being; 4. determine the genetic stock structure of naturally spawning GS from the Sacramento-San Joaquin river system; and 5. determine GS spawning site suitability and environmental requirements for specific life stages in the Feather River and provide GS adults, sub-adults, larvae and eggs tissue, and live specimens from the Sacramento system to UC Davis scientists conducting the studies outlined in the first four objectives. Information gained from these assessments will be valuable to decision-makers regarding environmental resource management options to restore ecological health and improve water management for beneficial uses of the Bay-Delta system.

2. Approach and Methods (Phase 1): Animal Sampling and Holding: Juvenile and subadult/adult GS will be collected primarily from the Feather and Sacramento Rivers using appropriate methods and gear (specific methods outlined in Task 5, below) and held at UC Davis. Fish will be transported to the UC Davis Aquatic Center in oxygenated plastic bags (juveniles) or tanks (subadults/adults) with river water that will be kept cool. Immediately upon arrival, fish will be transferred to 1-4-m diameter, fiberglass tanks with aeration and a continuous flow of unchlorinated, air-equilibrated well water. Tank temperatures will approximate river temperatures at the time of capture (except for specific experimental protocols, see below), and GS will be offered fish pellets *ad libitum* (except for specific experimental protocols, see below).

Phase 1 Tasks: Task 1, GS Temperature Tolerance Limits and Behavioral Tendencies and Swimming Performance: Temperature and current play important roles in the development and survival of young fish. Determination of GS acute temperature tolerance limits (critical thermal maxima and minima) will follow modifications of the Becker and Genoway (1979) method (10 replicates, 1°C increasing or decreasing temperature per 10 min), using the loss of equilibrium endpoint. Control fish will be subjected to the same protocol but without temperature changes. At the endpoint, two stopcocks will be immediately switched, flushing the vessel with ambient water to quickly recover the fish. Loss of equilibrium in fish indicates physical disorganization due to the experimental variable and loss of the fish's ability to escape from conditions leading to its death (Becker and Genoway 1979). Horizontal, temperature gradient tanks (1.5 m long) will be used for GS' (acclimated to 11, 15, or 19°C) behavioral tendencies (temperature selection) experiments. Water depth will be set to avoid vertical temperature stratification of the water, yet avoid alarming the test fish. Fresh water (10°C and 25°C) will continuously flow into opposite ends of the gradient tank, and a telethermometer/probes system and angled mirrors will allow observations (every 5 min during the 1-h experimental period) of individual GS' temperature selections without disturbance. Controls will use ambient temperature water flows from both ends of the tank. GS critical swimming velocities (U_{crit}) will be determined at 11, 15, and 19°C using a modified Brett-type

recirculating water flume incorporating a variable-speed motor (Brett 1964). Juvenile fish will be placed in the swimming chamber of the 9-l flume and, after a 60-min acclimation period, critical swimming velocity will be measured by step increases of 10 cm/s in water velocity at 10 min intervals starting at 10 cm/s until the fish is fatigued (Beamish 1978, Young and Cech 1996). Appropriate statistical models (ANOVA, Kruskal-Wallis, and post-hoc tests) will be used to compare GS size and acclimation temperature group means.

Task 2. Reproductive Characteristics of Wild GS: Sturgeon have low reproductive rates because of their late sexual maturity and long intervals between the consecutive spawnings. Reproductive characteristics and reproductive rates vary in different species and populations, and may be determining factors in their resilience to extinction under continuing fishery and partial loss of the spawning habitat. It is important to obtain baseline information (not yet known) on wild GS reproductive development and traits, as in our studies with Atlantic and white (WS) sturgeons (Van Eenennaam et al. 1996, Doroshov et al. 1997, Chapman et al. 1996). We will collect samples of gonads and pectoral finrays (along with body size and capture site data) from juvenile, subadult, and adult GS (30-60 fish) captured in the Sacramento River system, Klamath River, and in their estuaries. Gonadal tissue will be histologically processed and the slides examined to identify sex, stage of gametogenesis, and morphological characteristics of gonadal tissue and germ cells. Ovaries will be subsampled for the individual fecundity, oocyte size, and germinal vesicle migration (criterion of oocyte ripeness and closeness of female to spawning; Van Eenennaam et al. 1996, Doroshov et al. 1997). Age will be reconstructed from the analysis of finray sections.

Task 3. Assessment of Stress and its Impact on Reproduction: Stress negatively impacts the health, growth and reproductive success of fish. We will first characterize the quantitative and qualitative nature of the GS' stress response by defining their interrenal response and correlate this response to changes in reproductive hormones. GS will be maintained at the UC Davis Aquatic Center in outdoor tanks with continuous running water under natural light, permitting water quality and temperature monitoring, and continual supervision of trained staff. Prior to experimentation, GS gonadal tissue will be removed to determine sex and stage of reproductive development of each fish (see Task 2, above). To characterize the interrenal response of these fish, individual fish will be anesthetized with MS 222, and fitted with an indwelling cannula (caudal vein) for subsequent blood sampling. Blood samples will be analyzed for cortisol, testosterone, 17α , 20β DH-pregnenolone and estradiol using radioimmunoassay (Moberg et al., 1995; Faulkner and Moberg, 1997). We will also determine circadian cortisol secretion rhythms in both males and females.

Task 4. Genetic Analysis: We will use two molecular approaches (microsatellites and amplified fragment length polymorphisms, AFLPs) to address questions regarding the GS' genetic health (genetic variability) and the genetic integrity (population structure) in the Sacramento-San Joaquin basin. We will assess GS variability and develop species-specific markers to distinguish between GS and WS and to screen collected embryos. Recently, we found six of the eleven microsatellite loci to amplify well in GS (May et al. 1997). A very new technique (amplified fragment length polymorphisms, AFLPs) for rapidly screening large portions of the genome has recently become available (Vos et al. 1995), and we have successfully used AFLPs to identify subspecies of

endangered tui chubs (*Gila bicolor*), to build a linkage map for tilapia (*Oreochromis* and *Serathodon*), and to identify differences among populations of *Myxobolus cerebralis*, the parasite which causes whirling disease in fish. This approach leads to a variable number of bands per individual (with a target of 10-50 bands) depending primarily on the size of the genome and the number of base extensions used per primer. Variation is usually scored as presence or absence of bands; however, in many cases variation can be noted as sequence length differences. Whole embryos or fin samples will be collected and stored in 95% ethanol. Fin sections are taken with utility razor blades and these are exchanged and dissection boards rinsed with water and 70% ethanol between individuals. Genomic DNA will be extracted using the CTAB phenol/chloroform protocol (Saghai-Marooof et al. 1984, Doyle and Doyle 1987, Grewe et al., 1993).

Task 5. Determination of Sturgeon Spawning Habitats and Their Environmental Conditions:

Insulated aeration chambers will be constructed for transport of eggs and larvae collected from the Feather River. Artificial substrate will be fished continuously from March through June at 6 sites between Shanghai Bend (Rkm 41) and Thermalito outfall (Rkm 95) which have depth, velocity and substrate characteristics typical of spawning sites of other sturgeon. Substrates will be retrieved twice weekly, all eggs will be either preserved in fixatives compatible with later DNA analysis (microsatellite and AFLPs) to determine species (WS or GS) or will be transported alive to UC Davis for growout for species determination. Preserved embryo samples will be aged to back-calculate time of spawning using temperature-modified WS development times (Wang et al. 1985, Beer 1981) until species-specific information is developed from our captive breeding and culture studies. Twice weekly, larval nets (Kohlhorst 1976) will be fished at locations between the southern end of the Oroville Wildlife area (Rkm 87) and the Highway 99 bridge near Nicklaus (Rkm 15). Larvae will be either preserved or maintained alive for transport to UC Davis. Time of spawning will be estimated by WS larval development times (Beer 1981). Throughout the spawning seasons, flow will be monitored hourly by the CDWR at established flow recording stations immediately below the Thermalito outfall (Rkm 95), near the Gridley bridge (Rkm 81), near Yuba City (Rkm 45) and in the Yuba River near Marysville. This study will establish hourly temperature recording stations above the Thermalito outfall (Rkm 96), Gridley (Rkm 91), below the Yuba River, and near Nicklaus (Rkm 15) using submersible data loggers that will be weekly interrogated during late winter/spring. During deployment of artificial substrates, water velocity 30 cm above the substrate will be measured (current meter), bottom (substrate) samples will be taken with an Ekman dredge, and substrates will be indexed (Instream Flow Suitability Method). In San Pablo Bay and the west Delta, CDFG sturgeon gill-netting procedures will be modified and a live tank constructed to maintain captured GS alive for transport to UC Davis. We will also facilitate/coordinate the storage of GS juveniles captured at the John Skinner Fish Facility of the State Water Project for later transport to UC Davis.

Approach and Methods, (Phase 2): GS will be sampled and held as in Phase 1 (above).

Task 1. Food Consumption Rate, Growth Rate, and Respiratory Metabolism Measurements:

Juvenile GS, either from river and Delta collections or from captive breeding experiments (see below) will be used to assess temperature's effects on three critical functions: food consumption, growth, and metabolism. Food consumption rate and growth rate studies will be conducted

simultaneously on juvenile GS in replicate tanks at three temperature treatments: 11, 15, and 19°C (Myrick and Cech 1996). Fish will be situated in groups of 30 in five, replicate 110-L round fiberglass tanks (with continuous water and air flows) per temperature treatment. Fish will be fed Biodiet fish pellets twice daily, and uneaten pellets will be siphoned (and counted) twice daily to calculate food consumption rates (in g food/tank/day, with appropriate statistical comparisons between groups). Fish will be weighed and measured at the start and end of the 30-d experiment to determine growth rates, using appropriate statistical comparisons. Specific growth rate will be determined (Busacker et al. 1990) for comparisons with literature values on juvenile WS (Cech et al. 1984, Crocker and Cech 1996). Respiratory metabolic (oxygen consumption) rate measurements will be conducted on ten (post-food consumption and growth rates experiment) fish from each of the three temperature treatment groups (11, 15, 19°C) in closed respirometers following the methods of Cech (1990). If GS show significant activity in respirometers, experiments will be videotaped to quantify activity (Crocker and Cech 1997) and oxygen (convertible to energy units) costs of activity will be estimated (with appropriate statistics).

Task 2. Captive Breeding, Culture, and Characterization of Early Developmental Stages: GS captive breeding will provide critical material for our assessments, in addition to experience and techniques that may be needed for artificial reproduction of this rare species. We will collect 2-3 female and 3-5 male broodfish from the Sacramento and Klamath systems (gill nets or angling) and transport them by truck (with special oxygenated tank) to UC Davis. Broodfish will be held in 4-m diameter outdoor tanks with continuous water flows. Spawning and hatchery techniques will generally follow standard WS procedures (Conte et al. 1988, Van Eenennaam et al. 1996, Moberg and Doroshov 1996). Larvae and juveniles will be raised, at low density in smaller flow-through tanks, on the artificial (Biodiet) and/or natural (brine shrimp nauplii and tubifex) diets. Young GS survival, growth, feeding, and health will be maintained, and water quality will be monitored. We expect success, because Asian GS have large yolky eggs, and large and robust larvae at the onset of exogenous feeding (Artyukhin and Andronov, 1990), in contrast with lake and Atlantic sturgeon that possess more technically challenging larvae. We will incubate fertilized GS eggs (petri dishes or glass trays) in temperature-controlled (four temperatures: range 8-18°C) flow-through tanks (or in hatching jars at 10-20°C) and monitor development rates (including mortalities and abnormalities) using photomicrography and regression analysis (Wang et al. 1985, Dettlaff et al. 1993). Larval measurements and weights will yield temperature effects data on larval growth. Photomicrographs of embryos and larvae will be scanned, processed (Adobe Photoshop software) and compared with WS. Dettlaff et al. (1993) noted that species-specific differences in sturgeon are usually subtle during the embryo development, and different species are usually distinguished by the egg size and pigmentation patterns; however, the differences in morphology become prominent in larval stages, particularly before the transition to exogenous feeding (Wang et al. 1985).

Task 3. Responses to Stressors: GS' responses to stressors will be determined via brief air exposures in a dip net (simulating culture procedures) and ACTH₁₋₂₄ administrations (via the vascular cannula) to determine the maximum and temporal characteristics of the GS' interrenal response. At this time and during subsequent studies, we will monitor the effect of the interrenal response on gonadal steroid secretion. These data will be used to establish appropriate culture

conditions (e.g., tank size, stocking densities, handling practices, water temperature), should GS captive breeding/mitigative stocking be needed.

Task 4, Delta GS Stock Identification: The degree of reproductive isolation of Sacramento-San Joaquin GS from Klamath River and Rogue River (Oregon) populations will be determined using the microsatellites and AFLP DNA techniques employed in Phase I studies. Klamath River collections (coordinated with Tom Kisanuki [USFWS], Mike Orcutt [Hoopa Tribe] and Tony Fletcher [Yurok Tribe]) and Rogue River (coordinated with ODFW biologists) will quantitatively describe the regional uniqueness of Sacramento-San Joaquin GS.

Task 5, Determination of Sturgeon Spawning Habitats and Their Environmental Conditions: GS sampling and spawning habitat characterization will continue using the equipment and techniques that proved most successful during Phase 1 efforts.

b. Proposed Scope of Work: In this document, we describe two phases of a coordinated, CDFG-UC Davis multi-phase biological assessment of GS in the Sacramento-San Joaquin watershed. Phases 1 and 2 will be conducted during the first two project years, respectively. The Project Description and Approach (above) identifies the five tasks to be completed in each of these two phases. Tasks' deliverables and costs (Year 1) are: 1. juvenile GS' temperature tolerance limits and behavioral tendencies, and swimming performance (\$27,938); 2. wild GS' reproductive characteristics (\$53,370); 3. GS' baseline reproductive and stress hormone profiles (\$30,438); 4. genetically differentiation between GS and WS (\$43,471); and 5. identification of Feather River GS' spawning sites (\$41,321), Total for Year 1: \$199,046. Tasks' deliverables and costs (Year 2) are: 1. temperature effects on juvenile GS food consumption, growth, and metabolism (\$30,438); 2. captive breeding, culture, and characterization of GS' early developmental stages (\$48,370); 3. GS' responses to stressors (\$30,438); 4. genetic characterization of Sacramento-San Joaquin GS stock (\$48,471); and 5. identification of suitable river conditions for GS' spawning and larval rearing (\$40,979), Total for Year 2: \$198,696. The linked components of this study are not easily separated, because the individual tasks rely on deliverables from the others.

c. Location of Project: Project location is primarily in the Feather and Sacramento River systems, with additional work in San Pablo Bay, the John Skinner Fish Facility, and at UC Davis.

d. Expected Benefits: The GS is a CALFED priority (tier one) species, and the proposed assessments will focus on the biological characteristics of this species and its habitats towards their eventual restoration. We aim to provide the first information on spawning habitat requirements of green sturgeon and provide information on the status of all sturgeon spawning in the Feather River. Temperature sensors located above and below the Thermalito outfall will provide hourly temperature information during winter and spring periods that can be compared with hourly flow information currently available and begin to document temperature effects due to Thermalito afterbay operations in the lower Feather River. Assessments regarding GS' status, biological requirements (including temperature-related effects) in the Sacramento-San Joaquin watershed, and the feasibility of GS culture for future mitigational considerations may suggest how existing flow

regulation facilities (Oroville Dam, Thermalito Diversion Dam, water elevation and residence time in Thermalito afterbay) may be best employed to provide optimal flow and temperature conditions for sturgeon spawning. Collected information will significantly aid decision-makers regarding environmental resource management options to restore ecological health and improve water management for beneficial uses of the Bay-Delta system.

e. Background and Justification: Basic GS life history information is critical to this species' protection. Environmental requirements data are quantitatively linked via bioenergetic models (Jobling 1994) that allow predictions of physiological shortcomings (e.g., reduced growth, reproduction, and survival) associated with environmental stresses (measured by tolerance limits and hormonal responses) that lead to populational declines (Wedemeyer et al. 1990). American GS are known to spawn in the Sacramento and Klamath Rivers (Moyle et al., 1994) and the adults are present in the lower reaches of the Columbia and Fraser Rivers (Houston, 1988) but the spawning grounds, timing of spawning migrations, and developmental biology of this species are unknown. Similarly, no information exists on the GS' reproductive characteristics, such as gonadal development, age and body size at sexual maturity, fecundity, and egg size. Artyukhin and Andronov (1990) described spawning runs of the Asian GS (considered the same species *A. medirostris* Ayres or, as subspecies *A. medirostris mikadoi* Hilgendorf) in the Tummin River (Sakhalin Island) and succeeded in the artificial spawning of two females. However, they provided no detailed descriptions of early GS development. Stress negatively impacts the health, growth and reproductive success of fish. The GS' stress response (e.g., to temperature changes, low water quality) has never been studied and we have no knowledge regarding which natural or culture conditions are best for GS. Stress responses can lower reproductive success and, as a result, may account for unexplained failure of populations to reproduce normally. However, studies to test this hypothesis are not possible until a fundamental understanding of the GS' stress response is developed. Regarding identification of GS populations, the two primary advantages of AFLPs include the ability to randomly screen a large proportion of the genome and repeatability, thus scanning far more of the genome per unit of effort and cost than any other molecular genetic approach currently available. Our coordinated approach will significantly assist in GS recovery, a specific ERPP objective (Vol. 1, White and Green Sturgeon, pp. 146-148; Vol. 2, Feather River-Sutter Basin Ecological Zone, White and Green Sturgeon, p. 276).

f. Monitoring and Data Evaluation: This project encompasses monitoring/data evaluation using standard field, laboratory, and statistical techniques (see project description and approach, above).

g. Implementability: All necessary collecting permits (CDFG), animal care and use protocols (UC Davis), and cooperative arrangements (USFWS, Yurok and Hoopa Tribes) will be in place by the project start date (10-1-98). UC Davis has the appropriate laboratories and fish rearing facilities that will be required for this project.

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V. Costs and Schedule to Implement Proposed Project

a. Budget Costs

Table 1. Cost Breakdown of Funding Requested from the CALFED Program.

Project Phase & Task	Direct Salaries & Benefits	Supplies & Expenses	Equipment	Travel	Student Fee Remissions	In-direct Costs	Sub-contract	Total Cost
Phase 1 Task 1	15609	3727	0	2000	4468	2134	0	27938
Phase 1 Task 2	30560	8000	5000	1350	4468	3992	0	53370
Phase 1 Task 3	15609	8000	0	0	4468	2361	0	30438
Phase 1 Task 4	32919	6000	0	600	0	3952	0	43471
Phase 1 Task 5	See Table 2	See Table 2	See Table 2	See Table 2	See Table 2	2500	41329	43829
Phase 1 Totals	117602	34669	5000	6950	13404	14939	41329	199046
Phase 2 Task 1	15609	6000	0	2000	4468	2361		30438
Phase 2 Task 2	30560	8000	0	1350	4468	3992		48370
Phase 2 Task 3	15609	8000	0	0	4468	2361		30438
Phase 2 Task 4	32919	6000	5000	600	0	3952		48471
Phase 2 Task 5	See Table 2	See Table 2	See Table 2	See Table 2	See Table 2	0	40979	40979
Phase 2 Totals	118057	36192	5000	6950	13404	19093	40979	198696

Table 2. Subcontract Funding for CDFG (Task 5)

Project Phase	Direct Salaries & Benefits	Supplies and Expenses	Equipment	Travel	Indirect Costs	Total Cost
Phase 1	22905	8942	0	3000	6482	41329
Phase 2	23360	8192	0	3000	6427	40979

Table 3. Total (including "Leveraged") Funding for the Project.

Source of Funding	Phase 1 (Year 1)	Phase 2 (Year 2)	Funding Totals
UC Davis (matching)	18276	18276	36552
CDFG (in kind)	30000	30000	60000
CALFED (requested)	199046	198696	397692
Total	247322	246972	494294

Total Phase 1 funding requested from CALFED is \$199,046 for Phase 1 (first year) and \$198,696 for Phase 2 (second year). This amount (Tables 1, 2) includes funds for salaries and benefits of personnel (partial support of three graduate students, four undergraduate assistants, staff research associate, CDFG biologist, and associate research scientist), student fee remissions, equipment, supplies/expenses, travel, and overhead (@10% of non-student fee remissions and non-equipment costs for UC Davis and 18.6% of all costs for CDFG). The 10% (\$2,500) overhead on the CDFG subcontract is required only for the first \$25,000 of the subcontracted total (Table 1). The graduate student research assistants and undergraduate student assistants will assist in all aspects of the field and laboratory work, and the other staff will conduct the assessments and supervise/train the student assistants. Investigator B.P. May will require two months salary/benefits support. Student fee remissions are required in all UC Davis grants and contracts incorporating graduate student research assistants. Supplies and expenses include egg and larval aquaculture systems supplies, field sampling supplies, water and feed costs, histological supplies, film and developing, molecular biology supplies, chemicals and reagents, steroid analyses supplies, and physiological measurements supplies. Equipment needs include an aluminum fish transport tank (7'x4'x3', with 3" insulation and a 20"x18" dump gate @\$5,000, to be attached to the existing Aquaculture and Fisheries Program trailer) for Phase 1 and a thermocycler (polymerase chain reaction instrument to amplify regions of interest, such as microsatellite DNA, @\$5,000) for Phase 2. Travel costs are for truck/car rental and mileage for round trips to the Delta, Feather River, Sacramento River, Klamath River (with hotel and per diem costs) and to workshops/meetings to present/discuss results and implementation. "Leveraged" support (\$96,552 for Phases 1 and 2) is calculated from the 5% time commitment (salaries + benefits) of the faculty investigators for the project and the approximate \$30,000 in kind support by using CDFG personnel and vessels conducting white

sturgeon (WS) research funded by Federal Sport Fisheries Restoration Funds and matching state funds for obtaining GS, and by using State Water Project facilities and personnel to collect juveniles at the Byron fish screens (Table 3).

b. Schedule Milestones (for Phase 1):

September-November: (Initiation of CALFED Phase 1 funding) Initiate temperature tolerance, temperature selection experiments, and genetic tests for species identification; collection of gonadal samples (field trips to Delta).

December-February: First quarterly report; initiate swimming performance experiments and hormonal measurements; gonad sample processing and data analysis.

March-June: Second quarterly report; collection of eggs, larvae, and juveniles; characterize reproductive development, capture of broodfish, hatchery spawning, and experimental observations on early development; third quarterly report; continue experiments.

July - September: Continue studies, data analysis, first culture of juveniles, and preparation of annual report.

Phase 2 Milestones:

September-November: (Initiation of CALFED Phase 2 funding) Initiate food consumption and growth experiments, and genetic tests for stock identification; collection of gonadal samples (field trips to Delta).

December-February: First quarterly report; initiate metabolism experiments and stress response measurements; gonad sample processing and data analysis.

March-June: Second quarterly report; collection of eggs, larvae, and juveniles, capture of broodfish, hatchery spawning, and experimental observations on early development; third quarterly report; continue experiments.

July - September: Data analysis, culture of juveniles, and preparation of annual report. This annual report would be the final report if further phases of this GS project are not approved for funding.

c. Third Party Impacts:

Potential third party impacts resulting from this project include enhanced sport and native Californian fisheries resulting from improved fish populations. Also, California water consumers may benefit through restoration of a native, CALFED priority species, obviating its potential listing as a threatened or endangered species and restricting future water management options.

VI. Applicant Qualifications:

JOSEPH J. CECH, JR.

EDUCATION

B.S. U. Wisconsin, Madison, 1966 (Zoology); M.A. U. Texas, Austin, 1970 (Zoology); Ph.D. U. Texas, Austin, 1973 (Zoology)

POSITIONS

Resident Zoologist, Sea Search I, R/V Dante Deo, Caribbean Sea and S. Pacific Ocean, 1965-66; Research Assistant, U. Texas Marine Science Institute, 1966, 1968-72; Teaching Assistant, U.

Texas, 1967; Research Associate U. Texas Marine Science Institute, 1973; Research Associate, The Research Inst. Gulf of Maine, 1973-1975; Lecturer, U. Maine at Portland-Gorham, 1975; Assistant Professor 1975-1981, Associate Professor 1981-1987, Professor of Fisheries Biology, UC Davis, 1987-present; Associate Editor, *Transactions of the American Fisheries Society*, 1991-1993; Chair, UC Davis Dept. Wildlife, Fish, and Conservation Biology, 1992-1997.

FIVE SELECTED PUBLICATIONS

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SERGE I. DOROSHOV

EDUCATION

B.S. & M.S. (Zoology): U. Moscow, Russia, 1959; Ph.D. (Biology): Institute of Oceanology, Academy of Science, Russia, 1967.

POSITIONS

Fish Biologist, Inst. Freshwater Reservoirs, Acad. Sciences, Borok, Russia: 1959 - 1960; Research Scientist, Inst. Marine Fisheries and Oceanography, Moscow, Russia: 1961 - 1967; Head, Dept. Marine Aquaculture, Inst. Marine Fisheries and Oceanography, Moscow: 1969 - 1975; Aquaculture Expert, FAO, UN, Rome: 1975 - 1977; Visiting Professor, School of Fisheries, U. Washington, Seattle: 1977; Associate Professor 1978 - 1982; Professor, Dept. Animal Science, UC Davis: 1983 - present; Director, Aquaculture and Fisheries Program, UC Davis: 1995 - present.

FIVE RECENT PUBLICATIONS

1. Chapman, F.A., J.P. Van Eenennaam, and S.I. Doroshov. 1996. The reproductive condition of white sturgeon, *Acipenser transmontanus*, in San Francisco Bay, California. *Fish. Bull.* 94:628-634. 2. Van Eenennaam, J.P., S.I. Doroshov and G.P. Moberg. 1996. Spawning and reproductive performance of domestic white sturgeon (*Acipenser transmontanus*). In: S. Doroshov, F. Binkowski, T. Thuemler, D. MacKinlay (eds), *Culture and Management of Sturgeon and Paddlefish* (Symp. Proceedings, International Congress on the Biology of Fishes, San Francisco). pp. 117-122. 3. Van Eenennaam, J.P., S.I. Doroshov, G.P. Moberg, J.G. Watson, D.S. Moore and J. Linares. 1996. Reproductive conditions of the Atlantic sturgeon (*Acipenser oxyrinchus*) in the Hudson River. *Estuaries* 19(4):769-777. 4. Van Eenennaam, A., J.P. Van Eenennaam, J.F. Medrano, and S.I. Doroshov. 1996. Rapid identification of meiotic gynogenesis and polyploidy in white sturgeon (*Acipenser transmontanus* Richardson). *Aquaculture* 147:177-189. 5. Doroshov, S.I., G.P. Moberg and J.P. Van Eenennaam. 1997. Observations on the reproductive cycle of cultured white sturgeon, *Acipenser transmontanus*. *Env. Biol. Fish.* 48:265-278.

BERNARD (BERNIE) PAUL MAY

EDUCATION

B.S. U. Washington, 1973 (Molecular Biology); M.S. U. Washington, 1975 (Fisheries); Ph.D. Pennsylvania State U. 1980 (Genetics).

CURRENT POSITIONS

1995-Present Associate Research Biologist IV and Director, Genomic Variation Laboratory, Dept. Animal Science, UC Davis (75%); 1988-Present Senior Research Associate and Director, CLEEG, Department of Natural Resources, Cornell U. (25%); Assoc. Editor: Journal of Heredity

FIVE SELECTED PUBLICATIONS

1. Marsden, J.E., A. Spidle, and B. May. 1996. Review of genetic studies of *Dreissena* spp. Amer. Zool. 36:259-270. 2. Legge, J.T., R. Roush, B. May, R. DeSalle, and A. Vogler. 1996. Genetic criteria for establishing evolutionarily significant units in Cryan's buckmoth. Cons. Biol. 10(1):85-98. 3. May, B., C.C. Krueger, and H.L. Kincaid. 1997. Genetic variation at microsatellite loci in sturgeon: primer sequence homology in *Acipenser* and *Scaphirhynchus*. Can. J. Fish. Aquat. Sci. 54: 1542-1547. 4. May, B., T.A. Gavin, P.W. Sherman, and T.M. Korves. 1997. Characterization of microsatellite loci in the Northern Idaho ground squirrel, *Spermophilus brunneus* Mol. Ecol. 6:399-400. 5. May, B. 1998. Starch gel electrophoresis of allozymes. In: Molecular Genetic Analysis of Populations: A Practical Approach. 2nd Ed. A.R. Hoelzel, ed. Oxford Univ. Press.

GARY P. MOBERG

EDUCATION

B.A. Monmouth College, 1963 (Biology); M.S. U. Illinois, 1965 (Physiology); Ph.D. U. Illinois (Physiology); Postdoctoral Fellow, UC San Francisco 1970 (Neuroendocrinology)

RECENT ACADEMIC POSITIONS

Professor of Animal Science and Professor of Animal Physiology/Neurobiology, Physiology, and Behavior, 1982-present, UC Davis; Director of Aquaculture and Fisheries Program, 1991-93, UC Davis; Associate Dean for Animal Biology, College of Agricultural and Environmental Science, 1993-present, UC Davis.

FIVE SELECTED PUBLICATIONS

1. Van Eenennaam, J.P., S.I. Doroshov, G.P. Moberg, J.G. Watson, D.S. Moore and J. Linares. 1996. Reproductive conditions of the atlantic sturgeon (*Acipenser oxyrinchus*) broodstock in the Hudson River. Estuaries 19:769-777. 2. Doroshov, Serge I., Gary P. Moberg and Joel P. Van Eenennaam. 1997. Observations on the reproductive cycle of cultured white sturgeon (*Acipenser transmontanus*). Environmental Biology of Fishes. 48:265-278. 3. Pavlick, Raymond J., Jr. and Gary P. Moberg. 1997. Dopaminergic influence on gonadotropin secretion in white sturgeon. (*Acipenser transmontanus*). Fish Physiol. Biochem. 16:35-43. 4. Pavlick, Raymond J. Jr. and Gary P. Moberg. 1997. The effect of chronic testosterone administration on sturgeon gonadotropins in juvenile and pre-vitellogenic white sturgeon (*Acipenser transmontanus*). Gen.Comp. Endocr. 105:218-227. 5. Faulkner, Iwalani N. and Gary P. Moberg. 1997. Effects of short term management stress on the ability of GnRHa to induce gonadotropin secretion in male white sturgeon, *Acipenser transmontanus*. Aquaculture. 159:159-168.

RAYMOND G. SCHAFFTER

EDUCATION

B.S. U. Florida, Gainesville, 1965 (Zoology); M.S. U. Florida, Gainesville, 1968 (Botany).

POSITIONS

Instructor, Lake City Junior College, Lake City Florida, 1969-1971; Biologist, California Dept. Fish and Game 1973-present.

FIVE SELECTED PUBLICATIONS

1. Schaffter, R. G. 1980. Fish occurrence, size and distribution in the Sacramento River near Hood, California during 1973 and 1974. CDFG, Anadromous Fisheries Branch Report No 80-3. 2. Schaffter, R. G., P. A. Jones, and J. G. Karlton. 1983. Sacramento River and tributaries bank protection and erosion control report. CDFG. Sacramento, CA. 3. Schaffter, R. G. 1997. White sturgeon spawning migrations and location of spawning habitat in the Sacramento River. *Calif. Fish Game* 83:1-20. 4. Schaffter, R. G. 1997. Growth of white catfish in California's Sacramento-San Joaquin Delta. *Calif. Fish Game* 84:57-67. 5. Schaffter, R. G. 1997. Mortality rates of white catfish in California's Sacramento-San Joaquin Delta. *Calif. Fish Game* 84:45-56.

DAVID W. KOHLHORST

EDUCATION

B.S. U. Wyoming, Laramie, 1966 (Wildlife Conservation and Biology), M.A. UC Davis, 1970 (Zoology)

POSITIONS

Game Biologist, Washington Dept. Game, Olympia, 1971-1972; Junior Aquatic Biologist, 1972-1974; Assistant Biologist, 1974-1977; Associate Biologist, 1977-1995; and Senior Biologist (Specialist), California Dept. Fish and Game, Stockton, 1995-present.

FIVE SELECTED PUBLICATIONS

1. Kohlhorst, D.W. 1979. Effect of first pectoral fin ray removal on survival and estimated harvest rate of white sturgeon in the Sacramento-San Joaquin Estuary. *Calif. Fish Game* 65:173-177. 2. Kohlhorst, D.W. 1980. Recent trends in the white sturgeon population in California's Sacramento-San Joaquin Estuary. *Calif. Fish and Game* 66:210-219. 3. Kohlhorst, D.W., L.W. Miller, and J.J. Orsi. 1980. Age and growth of white sturgeon collected in the Sacramento-San Joaquin Estuary, California, 1965-1970 and 1973-1976. *Calif. Fish Game* 66:83-95. 4. Kohlhorst, D.W., L.W. Botsford, J.S. Brennan, and G.M. Cailliet. 1991. Aspects of the structure and dynamics of an exploited central California population of white sturgeon (*Acipenser transmontanus*). Pages 277-292 in: P. Willoit, editor. *Acipenser: First International Symp. on the Sturgeon*. CEMAGREF, Bordeaux, France. 5. Stevens, D.E., D.W. Kohlhorst, L.W. Miller, and D.W. Kelley. 1985. The decline of striped bass in the Sacramento-San Joaquin Estuary, California. *Trans. Am. Fish. Soc.* 114:12-30.

VII. Compliance with Standard Terms and Conditions:

The University of California, Davis, and the California Department of Fish and Game are public organizations of the State of California. Both organizations comply with the standard terms and conditions of non-discrimination and non-collusion. There are no conflicts of interest.